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This publication reports research involving pesticides. It does not contain recommendations for their use, nor does it imply that the uses discussed here have been registered. All uses of pesticides must be registered by appropriate State and/or Federal agencies before they can be recommended.

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# USE OF BIPHENYL FOR REDUCING PENICILLIUM DECAY OF STORED CITRUS

By L. G. Houck[1]

## SUMMARY

A test was conducted with 20 cartons each of lemons, navel oranges, and Marsh grapefruit to demonstrate the efficacy of biphenyl for controlling *Penicillium digitatum* decay, sporulation, and soilage. A portion of the fruit in each carton was inoculated and various dosages of biphenyl were evaluated.

Only the standard commercial treatment of two biphenyl sheets per carton provided adequate control of decay, sporulation, and soilage. Decay averaging 88 percent in nontreated lemons and 51 percent in navel oranges was reduced to 32 percent and 29 percent, respectively, in fruit receiving the standard biphenyl treatment. Sporulation was reduced from 100 percent in nontreated lemons, oranges, and grapefruit to 33, 36, and 19 percent, respectively, in the standard biphenyl-treated fruit. Soilage, likewise, was reduced from 85, 82, and 72 percent, for lemons, oranges, and grapefruit, to 29, 43, and 28 percent, respectively.

One biphenyl sheet per carton slightly reduced sporulation and soilage but had no effect on decay. Lesser dosages of one-half and one-quarter biphenyl sheet per carton were ineffective.

Biphenyl residues in fruit receiving the standard biphenyl treatment were highest in oranges, 89.1 parts per million (p.p.m.) on a fresh weight basis; intermediate in grapefruit, 30.9 p.p.m.; and lowest in lemons, 20.7 p.p.m.

## INTRODUCTION

Biphenyl[2] is used in the major citrus producing regions of the world to reduce decay and soilage of citrus fruit during postharvest shipping, storage, and marketing (*1*, *4*, *5*, *14*, *16*, *19*, *27*).[3]

Although decay control is important, prevention of soilage of sound fruit by the olive-green dusty spore masses from decayed fruit is equally as important. Heavy loads of fungus spores on sound fruit are unsightly and reduce the value of the fruit at the market. Also, these spores may increase decay of sound fruit later if fruit are injured during handling or improper storage. Biphenyl is valuable since it prevents or reduces fungus spore production.

The purpose of this paper is to show the relationship between biphenyl dosage, fruit decay, fruit soilage, and biphenyl residues in fruit.

The test described in this report was purposely conducted under conditions adverse for biphenyl effectiveness and optimum for absorption of biphenyl by fruit (*11*, *26*, *27*). Biphenyl absorbed by fruit does not control decay, mycelial growth, or sporulation of *Penicillium digitatum*. Biphenyl must be present in the free vapor form to be effective (*6*, *22*, *26*). More biphenyl is absorbed by citrus fruit stored at high temperatures; therefore, decay and soilage control by biphenyl is more effective at low temperatures (*11*, *27*).

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[2] Also called diphenyl.

[3] Italic numbers in parentheses refer to Literature Cited at the end of the report.

Therefore, in this test, fruit were stored at 22° C. (72° F.), not the recommended temperature for storing citrus fruit but a temperature at which fruit are sometimes held commercially, and a temperature at which biphenyl is less effective. This temperature is also near the optimum for *Penicillium digitatum* growth and development. Fruit were stored for 10 days. Longer storage would have masked the differences between biphenyl dosages used. Three varieties of citrus were included to demonstrate the differential absorption of biphenyl and decay control characteristics of different citrus varieties.

## LITERATURE REVIEW

The effectiveness of biphenyl for reducing fruit decay and fungus sporulation was reported first by Tompkins in England in 1936 (*31*). The commercial application of biphenyl has since been evaluated by many researchers in most citrus producing areas of the world.

Biphenyl is effective against *Penicillium digitatum* and *P. italicum*, the fungi causing green mold and blue mold decay, respectively, of citrus fruit. These are the two most important storage, shipping, and market diseases of California-Arizona citrus fruit. Biphenyl is also effective in reducing stem-end rots, caused by *Diplodia natalensis* and *Phomopsis citri*, and gray mold rot, caused by *Botrytis cinerea*, important decays in other citrus producing areas (*6*, *13*, *26*).

Biphenyl is a slightly volatile compound that vaporizes from the solid form at citrus storage temperatures and spreads throughout the atmosphere in the fruit containers. Biphenyl is a fungistat, rather than a fungicide (*3*, *6*, *26*,); that is, fungus growth and spore development are inhibited by biphenyl only so long as biphenyl is present at an effective concentration in the atmosphere. This material is relatively harmless to the fungus and once the biphenyl supply is exhausted or the fruit is removed from the biphenyl atmosphere, normal fungus growth and sporulation are resumed.

In the United States, biphenyl was tested in many shipments of oranges, lemons, and grapefruit by commercial firms during the late 1930's and early 1940's (*8*, *26*). In the California-Arizona citrus region, biphenyl has been tested in commercial fruit shipments since 1952. In 1956, the residue tolerance for biphenyl was established for oranges, lemons, and grapefruit (*17*). Since that time the use of biphenyl by the citrus industry has increased enormously.

Biphenyl was first used in impregnated tissue paper wrappers for individual fruit (*1*, *5*, *16*, *19*, *26*, *31*). In the United States, the practice of wrapping fruit was largely discontinued in the early 1950's when the wooden shipping box was replaced by the telescopic fiberboard carton. Biphenyl was then impregnated in cardboard liners or collars for citrus cartons, or in the cartons themselves (*9*, *10*). This method of application never became popular, and today most of the biphenyl used in the United States is added to kraft paper sheets, which are inserted in the citrus cartons at the time the fruit are packed. Biphenyl-impregnated fruit wrappers are still used in many countries where labor costs are low or where machine packing of fruit is not practiced.

Biphenyl, as used in the California-Arizona citrus industry, is normally applied to kraft paper at the rate of 1.8 to 2.3 kilograms per 92.9 square meters of paper (4 to 5 lb./1,000 sq. ft.). The paper is cut into sheets approximately 27 by 41 centimeters (10-9/16 by 16-1/2 in.) with six punched holes, 2.54 cm. (1 in.) in diameter. Biphenyl per standard sheet approximates 2.15 to 2.35 grams, or 4.3 to 4.7 grams biphenyl per carton (28). Two sheets are usually added per standard 28.19 liter (four-fifths bushel) fiberboard fruit carton, one at the bottom of the carton or one layer of fruit up from the bottom, the other at the top or one layer of fruit down from the top of the carton. Biphenyl is used with nearly all lemons, oranges, and grapefruit shipped from California and Arizona, whether for domestic consumption or export to foreign markets.

Biphenyl has sometimes been objected to because it imparts a characteristic odor to treated fruit (*7*, *25*, *31*). This odor, however, is detectable for only 2 or 3 days after fruit are removed from biphenyl (*25*, *30*, *31*). Even though

a persistent biphenyl residue remains in the peel of treated fruit, persons with acute sensitivity to biphenyl can not detect it 2 or 3 days after removal of the fruit from the carton (*30*). This characteristic odor has prevented biphenyl from being used on other fruits and vegetables.

In the United States, the legal maximum residue of biphenyl allowable in citrus fruit is 110 parts per million (p.p.m.), on a fresh fruit basis (*17*, *18*), whereas in countries of the European Economic Community it is 70 p.p.m. (*2*).

## PROCEDURES

*Fruit.*—Fruit furnished by Sunkist Growers Inc., Ontario, Calif., was obtained November 27 and 30, 1970, at packinghouses after normal commercial washing, waxing, and packing procedures, except that biphenyl sheets were omitted from the cartons during packing. These were added later as necessary for the test.

Sixty standard, vented (16 or 18 2.54-cm. holes per carton), four-fifths bushel, telescopic corrugated fiberboard cartons of fruit were used in the test. The fruit included 20 cartons each of lemons, navel oranges, and Marsh grapefruit. The lemons (average diameter 5.4 cm., 165 fruit per carton) were from the Upland area; the oranges (average diameter 6.6 cm., 113 fruit per carton) were from Strathmore in the Central Valley; and the grapefruit (average diameter 9.9 cm., 36 fruit per carton) were from the Imperial Valley. Shipping wax used on the grapefruit, but not on the lemons or oranges, contained a small amount of thiabendazole (TBZ), a fungicide commonly used on citrus fruit for postharvest soilage and decay control. TBZ has been cleared for use by the Food and Drug Administration and the USDA. A maximum residue of 2 p.p.m. for citrus fruit has been established. Normally, the particular concentration of TBZ used on these grapefruit is effective for controlling decay but not for reducing fungus sporulation.

*Inoculation.*—Since the natural decay rate of California-Arizona citrus fruit is usually quite low, some fruit in each carton were artificially inoculated so there would be an adequate number of decayed fruit for sporulation control and decay control observations. Of the fruit to be inoculated, half were severely inoculated by injecting 1/2 to 1 ml. of a *Penicillium digitatum* spore suspension just under the peel, using a syringe and hypodermic needle. All such heavily inoculated fruit decayed. Spores germinated and fungus mycelia grew, unaffected by biphenyl until they broke through the fruit surface. These hypodermically inoculated fruit were used to observe the effect of biphenyl upon fungus sporulation. The fruit for decay-control observations were inoculated by scratching fruit once (1 mm. deep by 1 cm. long) then brushing the scratch with a few drops of the *Penicillium digitatum* spore suspension. This type of inoculation caused 88 and 51 percent decay, respectively, of lemons and oranges not treated with biphenyl (table 1). Only three scratch-inoculated grapefruit decayed, probably because TBZ in the wax coating and the shallow 1-mm. scratch (a 2-mm.-deep scratch might have been more effective on these thick-skinned grapefruit) combined to prevent these grapefruit from decaying under the temperature and humidity conditions of this experiment.

*Penicillium digitatum* spores to be used for inoculation were cultured on enriched potato dextrose agar in petri dishes. The isolate used had been previously tested and found to be sensitive to biphenyl. This was necessary for this test, since there are biphenyl-tolerant strains of *Penicillium digitatum* (*3*, *8*). Spore suspensions for inoculation purposes were prepared by suspending *Penicillium digitatum* conidia in sterile distilled water to which 0.1 percent Triton X-100 was added as a dispersing agent. Concentration of the inoculum suspension was adjusted to approximately $1 \times 10^6$ conidia/ml.

*Biphenyl treatments.*—Standard biphenyl-impregnated sheets of kraft paper were obtained from a citrus packinghouse. These sheets were 26.5 by 40.5 cm. and had six holes (2.54 cm. in diameter) per sheet to allow air circulation in the cartons and rapid cooling during refrigeration.

For this text, normal commercial biphenyl dosage, no biphenyl, and three intermediate dosages were compared. Normal biphenyl dos-

age is two sheets per carton. One sheet was placed at the bottom and one sheet at the top of each carton receiving this treatment. Other treatments were one-half, one-quarter, and one-eighth normal biphenyl dosage, obtained by inserting 1, one-half, or one-quarter biphenyl sheet at the top of each carton receiving these treatments.

*Replication.*—Each of the treatments was replicated in four cartons of each of the three types of citrus fruit, 20 cartons of each variety, 60 cartons in all. In the test, 3,300 lemons, 2,260 oranges, and 720 grapefruit, or 6,280 fruit, were tested and inspected.

*Packing and storage.*—Predetermined numbers of inoculated and noninoculated fruit were counted and packed into citrus cartons. Lemons were jumble packed with inoculated fruit distributed at random throughout the carton. Oranges and grapefruit were handpacked in the 5-4-5-5 and 4-4-3-3 patterns, respectively. These are standard commercial packing patterns for these size fruit. Inoculated oranges and grapefruit were distributed equally among layers, but randomly within layers.

Cartons were stacked for storage, four cartons of the same treatment and fruit variety per stack, in a temperature-controlled room at 21° to 22° C. (70° to 72° F.). After 10 days, fruits were removed and inspected.

## OBSERVATIONS

*Decay.*—Decay control was determined by counting the healthy and decayed scratch-inoculated fruit. Miscellaneous or natural decays of noninoculated fruit were also counted, but there were not enough of these to provide meaningful data. A fruit was classified as "decayed" if only a small but definite fungus lesion was present or if the whole fruit was rotted. Preliminary observations did not show an apparent appreciable effect upon speed of decay development between treatments.

*Sporulation.*—Both hypodermically and scratch-inoculated fruit were used to observe the effect of the various biphenyl dosages upon *Penicillium digitatum* sporulation intensity. A visual estimate of average sporulation possible, as compared with the nontreated control, was made by a five-person panel, taking into account such factors as thickness and looseness of the spore load and amount of the fruit surface covered.

*Soilage.*—Sound, noninoculated fruit from each carton were separated into two categories, "soiled" fruit with any *Penicillium* spore dust or parts from decaying fruit, and "clean" fruit without *Penicillium* spores or decay material.

*Reduction index.*—The percent decay or soilage found by inspecting the three varieties of fruit was transformed into a "reduction index" (RI) following the formula: RI = (percent decay or percent soilage in control) - (percent in treatment) ÷ (percent in control) × 100. This index provides a convenient means of comparing the effectiveness of the different treatments between fruit varieties because the percent decay or soilage in control fruits varied between varieties.

*Biphenyl analyses.*—Biphenyl on or absorbed by fruit during storage was determined by thin-layer chromatography and spectrophotometric procedures according to the method described by Norman, Rygg, and Wells (*24*). Forty fruit were used for each analysis, 10 sound fruit selected at random from each of the four cartons in each treatment and fruit variety. Samples were collected at time of observation, fruits were immediately ground and an aliquot frozen for later analysis. Duplicate samples of 300 g. of the ground fruit were analyzed and the results were averaged. Biphenyl content is expressed as parts per million on a fresh fruit basis.

## RESULTS AND DISCUSSION

*Decay.*—Decay of scratch-inoculated lemons (table 1) was reduced from 88 percent in the untreated controls to 32 percent in the standard biphenyl treatment, an RI of 64. Less biphenyl did not appreciably reduce decay of scratch-inoculated lemons. Decay of navel oranges was also reduced but not as much. The standard treatment had an RI of 43 (from 51 percent decay in the scratch-inoculated control treatment to 29 percent decay in the standard biphenyl treatment). In this experiment, scratch-inoculated grapefruit did not decay, for reasons previously discussed. Average decay RI with lemons and oranges was 55 for the standard two biphenyl sheets per carton, whereas the treatment with one sheet per carton had an RI of 12.

All hypodermically inoculated fruit decayed, since biphenyl could not contact the fungus spores beneath the peel. Natural decay occurring in noninoculated fruit was very low, varying between 0 and 3 fruits per carton. No correlation was noted between biphenyl dosage and natural decays.

Had the storage temperature been lower, efficacy of biphenyl would have been greater (*7, 11, 27, 28*). However, even at this relatively high storage temperature, control of *Penicillium* decay was very adequate.

Decay of fruit is prevented by inhibition of

Table 1.—Decay control, sporulation, soilage, and biphenyl residues of lemons, navel oranges, and grapefruit stored 10 days at 21° to 22° C. with standard and reduced biphenyl dosages

| Biphenyl sheets per carton | Fruit per treatment[1] | Decay[2] | Sporulation[3] | Soilage[4] | Biphenyl residues[5] |
|---|---|---|---|---|---|
| | *Number* | *Percent* | *Percent* | *Percent* | *P.p.m.* |
| *Lemons* | | | | | |
| 0 | 676 | 88 | 100 | 85 | 2.4 |
| ¼ | 663 | 70 | 100 | 80 | 6.9 |
| ½ | 672 | 80 | 100 | 85 | 5.9 |
| 1 | 676 | 75 | 66 | 60 | 13.2 |
| 2 | 637 | 32 | 33 | 29 | 20.7 |
| *Navel oranges* | | | | | |
| 0 | 453 | 51 | 100 | 82 | 18.4 |
| ¼ | 452 | 50 | 99 | 87 | 50.0 |
| ½ | 452 | 45 | 97 | 86 | 48.9 |
| 1 | 452 | 47 | 90 | 63 | 67.7 |
| 2 | 453 | 29 | 36 | 43 | 89.1 |
| *Grapefruit* | | | | | |
| 0 | 144 | [6]4 | 100 | 72 | 7.7 |
| ¼ | 144 | 0 | 100 | 73 | 17.6 |
| ½ | 144 | 0 | 89 | 64 | 17.8 |
| 1 | 144 | 8 | 44 | 40 | 23.7 |
| 2 | 144 | 0 | 19 | 28 | 30.9 |

[1] Stored in 4 cartons.

[2] Decay of 100, 80, or 24 scratch-inoculated lemons, oranges or grapefruit, respectively, per treatment.

[3] Estimated by a 5-member panel, from observations of 200, 160, or 48 scratch- and hypodermically inoculated lemons, oranges, and grapefruit, respectively, per treatment.

[4] Soilage of up to 460, 292, or 96 sound, noninoculated lemons, oranges, and grapefruit, respectively, per treatment.

[5] Residues of a pooled sample of 40 fruit per treatment, 10 fruit selected at random from each of the 4 cartons per treatment.

[6] Scratch-inoculated grapefruit did not decay. Explanation in text.

germ tube development after spore germination (*3*). Fungus hyphae, as long as they are in a sufficiently biphenyl-enriched atmosphere, do not grow and spores, which might infect fruit during later handling and wounding after biphenyl is exhausted or removed, are not formed (*26*).

The well-proven combination of sodium o-phenylphenate (SOPP) and biphenyl still represents one of the best and cheapest decay controls available (*7*, *14*). The method is subject to abuse, however, if proper refrigeration is not used (*7*). There are reports of excessive decay in fruit shipped to distant markets in spite of the use of biphenyl (*29*).

*Sporulation.*—As biphenyl dosage increased, fungus sporulation decreased (table 1). The standard biphenyl dosage reduced sporulation 64, 67, and 81 percent on oranges, lemons, and grapefruit, respectively, an average of 71 percent reduction for all fruit. One biphenyl sheet per carton reduced sporulation an average of 33 percent for all three varieties, but lesser dosages of biphenyl were ineffective.

Fungus sporulation is also slowed by cooler temperatures, and the combination of biphenyl and refrigeration is more effective than either treatment alone (*15*, *28*). Biphenyl does not always prevent sporulation of molded fruit, however, (*19*) perhaps because of the occasional presence of biphenyl-tolerant strains of the fungus (*3*, *8*) or because biphenyl may not be continuously present in the storage atmosphere, as required for sporulation control.

*Soilage.*—The standard biphenyl dosage was more effective than the reduced dosages for limiting sporulation and the associated soilage of noninoculated fruit (table 1). Soilage of lemons was reduced from 85 to 29 percent, an RI of 66; grapefruit soilage was reduced from 72 to 28 percent, an RI of 61; and soilage of oranges was reduced from 82 to 43 percent, an RI of 48. One biphenyl sheet per carton did reduce soilage, with RI values of 18, 23, and 44 (average 29), respectively, for lemons, oranges, and grapefruit. Soilage of fruit receiving lesser biphenyl dosages was similar to soilage of nontreated fruit.

Biphenyl is more effective in reducing soilage than decay, especially at refrigerated temperatures (*29*). This is shown only to a slight degree at the relatively high temperature used in this experiment, the average decay RI of lemons and oranges being 53 and the average soilage RI of lemons and oranges being 57.

*Biphenyl residues.*—As biphenyl dosage increased so did biphenyl residues in the fruit (table 1). This agrees with the results of others (*9*, *12*, *21*). In this experiment, no biphenyl residues exceeded the United States tolerance of 110 p.p.m. (*17*, *18*), and most residues were far below this level. Navel oranges stored with two biphenyl sheets per carton absorbed 89.1 p.p.m. biphenyl, exceeding the European Economic Community tolerance of 70 p.p.m. (*2*), while grapefruit and lemons absorbed 30.9 and 20.7 p.p.m., respectively.

In this experiment, the storage room used was new and fairly airtight, and fruit without biphenyl sheets in the cartons absorbed biphenyl from the room atmosphere, as shown in table 1. This observation has been reported (*11*, *21*, *30*).

Factors affecting biphenyl residues in fruit have been identified as storage temperature, length of storage, biphenyl dosage, carton venting, fruit variety, and location or placement of biphenyl sheets in the cartons (*9*, *10*, *11*, *12*, *21*, *23*, *27*).

Although it was once suggested that absorbed biphenyl was effective in reducing decay (*12*), more recent work by Norman et al. (*22*) has shown fairly conclusively that absorbed biphenyl in navel oranges does not control decay, mycelial growth, or sporulation. They and others (*3*, *6*) have maintained that control is due to the amount of biphenyl vapor present in the atmosphere surrounding the fungus.

Biphenyl odor in relation to residues in fruit has been studied extensively (*25*, *30*). Biphenyl odor disappears rapidly from fruit, generally within a day or two after biphenyl is removed, although residues may persist for several weeks (*12*, *27*, *28*). The odor is due to the fraction of total biphenyl residue sorbed on the fruit surface. There is no relationship between the amount of biphenyl absorbed in fruit and odor; in fact, fruits containing large amounts of biphenyl generally had only a faint odor, whereas those having a low content of biphenyl had a strong odor (*30*).

*Varietal differences.*—Biphenyl control of decay was greater in lemons than in navel oranges (table 1), the decay RI for lemons being 64 and for oranges 43.

Navel oranges absorbed the most biphenyl, whereas grapefruit absorbed one-third as much and lemons 1/4 to 1/5 as much as the oranges. Harvey (*9*) also observed lemons absorb less biphenyl than either oranges or grapefruit. Hayward and Edwards (*11*) working with 14 varieties of citrus fruit found strikingly dissimilar biphenyl absorption in different varieties. Their reports (*11*, *12*,) show tangerines take up more biphenyl than other citrus fruit. Norman et al. (*20*) showed little difference in biphenyl absorption of navel and Valencia oranges. Hayward and Edwards (*11*) found Duncan and Marsh grapefruit absorb nearly the same amounts of biphenyl.

Biphenyl decay control may be related to absorption of biphenyl by fruit, the more biphenyl the fruit absorb the less there might be in the atmosphere or the sooner the supply could be exhausted. To our knowledge, however, this possibility has not been demonstrated.

*Biphenyl dosage.*—In this test, only the commercial treatment of two biphenyl sheets per carton provided adequate decay, sporulation, and soilage control, although one sheet per carton did provide slight soilage and sporulation reduction (table 1).

Commercially, two biphenyl sheets containing 4 pounds biphenyl per 1,000 square feet of paper have long been considered to be the minimum practical dosage at which the material should be used, although the 2 pounds per 1,000 square foot rate does have some effect (*9*).

Cost of material and legal tolerance are other factors to be considered when determining effective dosages of biphenyl for citrus fruit (*28*).

## LITERATURE CITED


(1) Christ, R. A. 1964. The use of diphenyl as a protectant for citrus fruit shipped in export cases. South African Citrus Jour. 362: 5-11.

(2) Dormal-vanden Bruel, S. 1969. Pesticides in citrus fruits, and common market regulation and tolerances. First Internatl. Citrus Symposium Proc. 2: 1023-1026.

(3) Duran, R., and Norman, S. M. 1961. Differential sensitivity to biphenyl among strains of *Penicillium digitatum* Sacc. U.S. Agr. Res. Serv. Plant Dis. Rptr. 45: 475-480.

(4) Farkas, A. 1938. The practical application of impregnated wrappers against fungal decay of citrus fruits. Hadar 11: 261-267.

(5) _____ 1939. Control of wastage of citrus fruits by impregnating wrappers on a commercial scale. Hadar 12: 227-230.

(6) _____ and Aman, J. 1940. The action of diphenyl on *Penicillium* and *Diplodia* moulds. Palestine Jour. Bot. 2: 38-45.

(7) Grierson, W., Hayward, F. W., and Oberbacher, M. F. 1960. Simulated packing, shipping and marketing experiments with Valencia oranges. Fla. State Hort. Soc. Proc. 72: 248-254.

(8) Harding, P. R., and Savage, D. C. 1961. Biphenyl-resistant strains of citrus green mold. Calif. Citrog. 46: 280, 306-308.

(9) Harvey, E. M. 1955. Absorption of biphenyl from biphenyl-treated cartons by citrus fruits and its effect on decay, 1953-54. U.S. Dept. Agr. Agr. Market. Serv. AMS-3, 16 pp.

(10) _____ and Sinclair, W. B. 1953. Diphenyl—its uses in the citrus industry. A report compiled by a technical committee appointed by the Citrus Industry Association, 27 pp.

(11) Hayward, F. W., and Edwards, G. J. 1964. The effects of time and temperature of storage on residues of diphenyl in citrus fruits packed in cartons with diphenyl pads. Fla. State Hort. Soc. Proc. 76: 318-320.

(12) _____ and Edwards, G. J. 1965. Some factors affecting the level and persistence of diphenyl residues in citrus fruit. Fla. State Hort. Soc. Proc. 77: 315-318.

(13) Hopkins, E. F., and Loucks, K. W. 1947. The use of diphenyl in the control of stem-end rot and mold in citrus fruits. Citrus Indus. 28(10): 5-9.

(14) _____ and Loucks, K. W. 1950. Combination of Dowicide "A" with diphenyl for the control of decay

in citrus fruits. Citrus Mag. 12(11): 24-28.

(15) _____ Loucks, K. W. 1966. Value of Dowicide A - hexamine treatment in cold storage of oranges. Citrus Mag. 18(10): 16-20.

(16) Huelin, F. E. 1942. The handling and storage of Australian oranges, mandarins, and grapefruit. Austral. Council Sci. & Indus. Res. Bul. 154, 60 pp.

(17) Larrick, G. P. 1956. Tolerances for residues of diphenyl. Fed. Reg. 21: 5619-5620.

(18) _____ 1960. Tolerances for residues of biphenyl. Fed. Reg. 25: 13631-13632.

(19) Leggo, D., and Seberry, J. A. 1965. The effect of diphenyl wraps and case liners on green mould wastage in stored oranges. Austral. Jour. Expt. Agr. and Anim. Husb. 5: 91-96.

(20) Norman, S., Craft, C. C., and Fouse, D. C. 1971. Biphenyl absorption of citrus fruit: the effect of variety, color class, and injury by freezing, peeling, and lack of oxygen. U.S. Dept. Agr. Market. Res. Rpt. 896, 6 pp.

(21) _____ Craft, C. C., Fouse, D. C., and Rygg. G. L. 1969. Biphenyl residues, decay, and soilage in oranges: effect of carton venting, barriers, and biphenyl dosages during storage. U.S. Dept. Agr. Market. Res. Rpt. 853, 16 pp.

(22) _____ Fouse, D. C., and Craft, C. C. 1969. Comparison of absorbed biphenyl in oranges and biphenyl vapor in controlling decay, mycelial growth, and sporulation by *Penicillium digitatum*. U.S. Agr. Res. Serv. Plant Dis. Rpt. 53: 780-784.

(23) _____ Rygg, G. L., and Fouse, D. C. 1968. Biphenyl residues in oranges: effect of biphenyl dosage and pad placement. U.S. Dept. Agr. Agr. Res. Serv. ARS 51-26, 8 pp.

(24) Norman, S., Rygg, G. L., and Wells, A. W. 1966. Improved cleanup method for determination of biphenyl in citrus fruits and in biphenyl-impregnated kraft papers by thin layer chromatography. Assoc. Off. Anal. Chem. Jour. 49: 590-595.

(25) Rajzman, A. 1965. The odor of oranges protected by biphenyl-impregnated wraps and stored in wooden boxes. Rehovot Natl. Univ. Inst. Agr. Prelim. Rpt. 495, 24 pp.

(26) Ramsey, G. B., Smith, M. A., and Heiberg, B. C. 1944. Fungistatic action of diphenyl on citrus fruit pathogens. Bot. Gaz. 106: 74-83.

(27) Rygg, G. L. 1969. Tests of overseas shipments of California citrus fruit—interrelations between fruit condition, biphenyl content, transit temperature, delayed loading, and biphenyl pad placement. U.S. Dept. Agr. Market. Res. Rpt. 830, 22 pp.

(28) _____ Wells, A. W., Norman, S. M., and Atrops, E. P. 1962. Biphenyl control of lemon spoilage. U.S. Dept. Agr. Market. Res. Rpt. 569, 15 pp.

(29) _____ Wilson, C. W., and Garber, M. J. 1961. Effect of biphenyl treatment and carton ventilation on decay and soilage of California lemons in overseas shipments. U.S. Dept. Agr. Market. Res. Rpt. 500, 14 pp.

(30) Souci, S. W., and Maier-Haarlander, G. 1966. The diphenyl content and odour of citrus fruits and citrus products and the contamination by diphenyl of untreated fruits during storage (in German). Residue Rev. 16: 103-137.

(31) Tomkins, R. G. 1936. Wraps for the prevention of rotting of fruit. [Gt. Brit.] Dept. Sci. and Indus. Res., Food Invest. Bd. Rpt., pp. 129-131.